Optimizing vitrification method in *in vitro*-produced bovine blastocysts: Effect of the exposure temperature to cryoprotectants in their osmotic behavior

Universitat Autònoma de Barcelona

FACULTAT DE VETERINÀRIA

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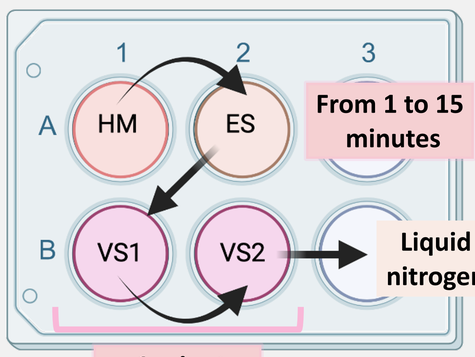
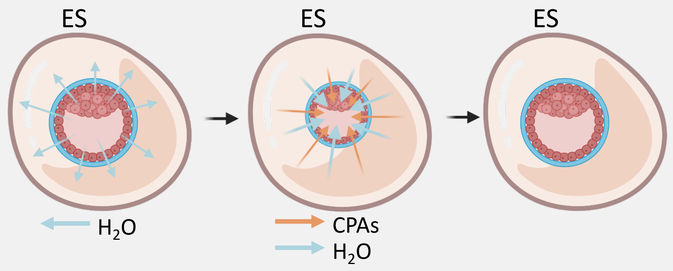
INTRODUCTION





↑ Temperature
↑ Exposure time
→ ↑ PERMEABILITY
→ ↑ TOXICITY

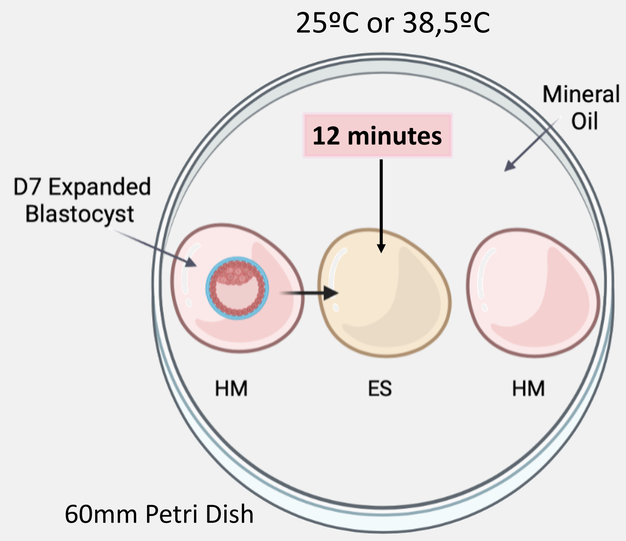
HM: Holding medium / ES: Equilibration solution / VS1 and VS2: Vitrification solution 1 and 2 / CPAs: Cryoprotectants

OBJECTIVE

To evaluate the **effect of temperature in the osmotic behavior** of *in vitro*-produced bovine blastocysts during the exposure to **the equilibration solution** comparing two temperatures: **25ºC and 38.5ºC**, in order to establish the **optimal exposure time** to each temperature.

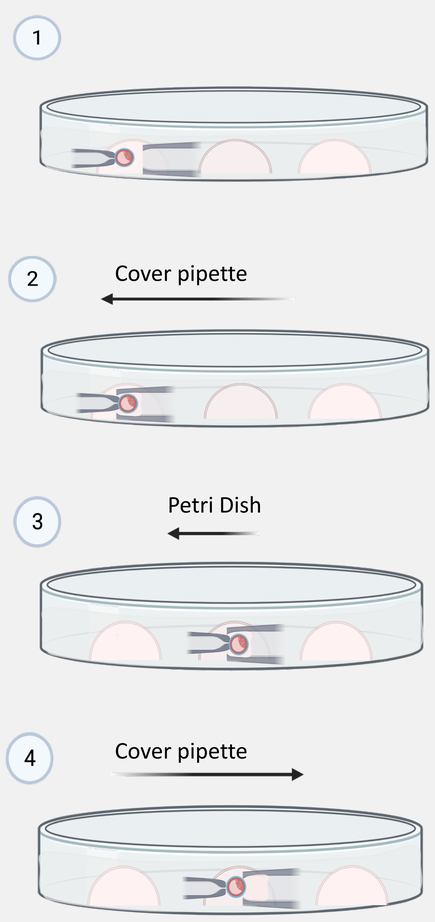
IMPROVE THE CURRENT VITRIFICATION METHOD

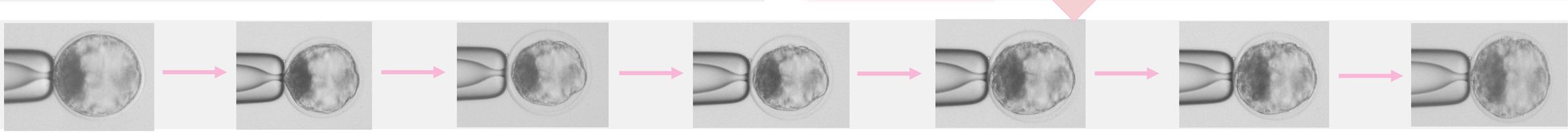
TIME LAPSE ANALYSIS



TIME-LAPSE CAPTURES

MEDIUM COMPOSITION:
HM: HEPES-TCM-199 + 20% Fetal calf serum
ES: HM + 7.5% ethylene glycol + 7.5% dimethylsulfoxide



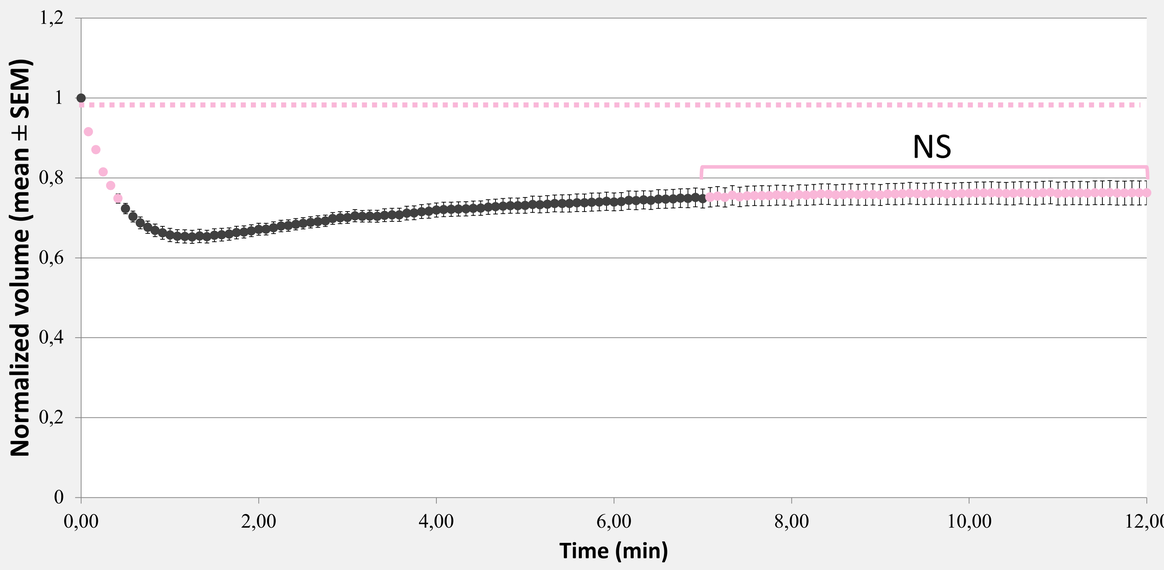


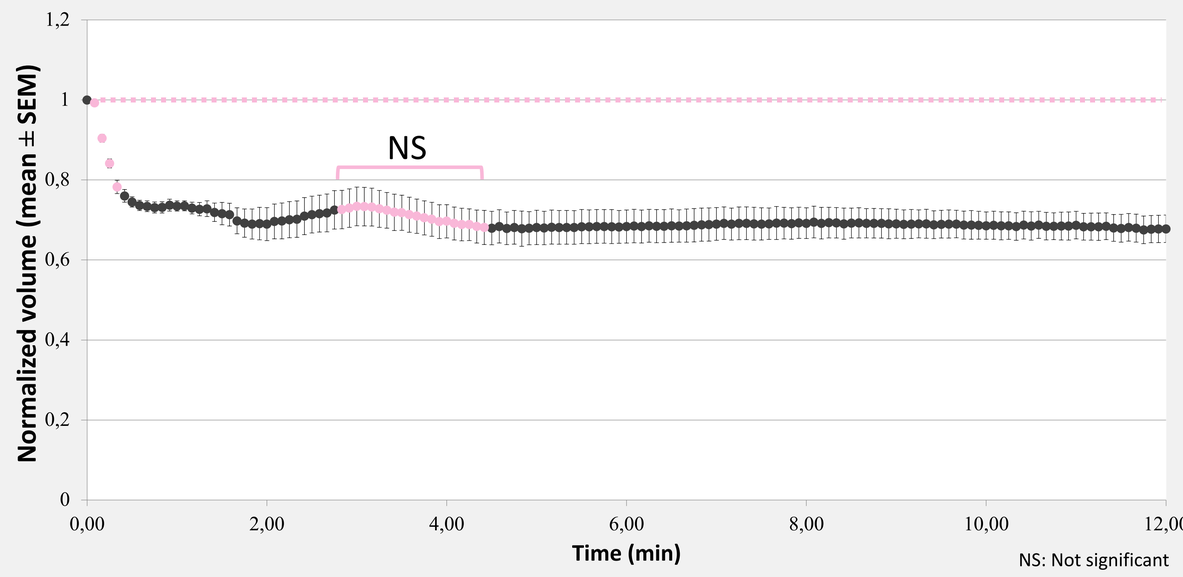
RESULTS 25ºC

Volume recovery	n	%
Over 80% of the initial volume	6	30%
Between 55% and 80% of the initial volume	10	50%
Less than 55% of the initial volume	4	20%

38.5ºC

Volume recovery	n	%
Over 80% of the initial volume	2	11%
Between 55% and 80% of the initial volume	8	42%
Less than 55% of the initial volume	9	47%





NS: Not significant

DISCUSSION

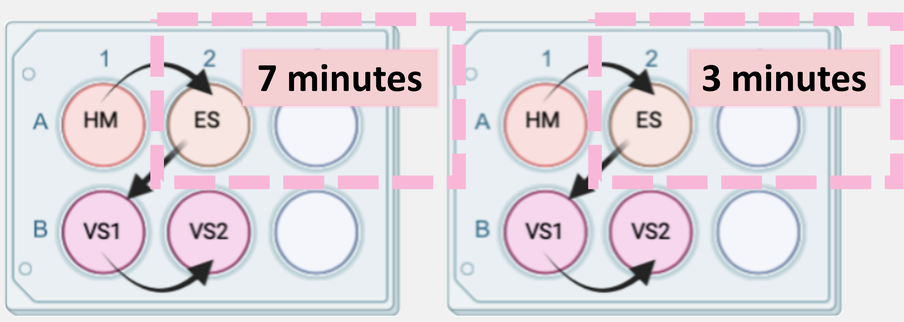
Temperature modifies the osmotic response of day 7 expanded blastocysts exposed to ES

Establish different incubation times depending on the exposure temperature to ES

OSMOTIC RESPONSE:
- 25ºC → Results comparable to those reported in previous studies
- 38.5ºC → Analyzed for the first time

CONCLUSIONS

25ºC — Shorter protocols — 38.5ºC



+ VITRIFICATION SURVIVAL OUTCOMES